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CHLORINATED MUNICIPAL WASTE TOXICITIES TO RAINBOW TROUT AND FATHEAD MINNOWS

Michigan Department of Natural Resources
Lansing, Michigan

October 1971

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Chlorinated Municipal Waste Toxicities to Rainbow Trout and Fathead Minnows

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16. Abstract
Effect of chlorinated municipal wastes on fish life and laboratory findings of earlier researchers were assessed in separate studies at four Michigan municipal wastewater treatment plants. Tolerance levels and river length below each plant outfall rendered unavailable to resident fish populations were determined. Chlorine in municipal wastewater discharges is usually present in combined form with ammonia, ammonium hydroxide or ammonium ions to form mono-, di-, or trichloramine. Ten rainbow trout (Salmo gairdneri) and ten fathead minnows (Pimephales promelas), previously acclimated, were held 96 hours in live boxes in the stream above and below these outfalls. Fish held below these outfalls were subjected to both chlorinated and non-chlorinated exposures during effluent discharge. Test waters were monitored chemically and bacteriologically during fish exposure. Total residual chlorine concentrations below three plants were toxic to rainbow trout at distances up to 0.8 mile. Fathead minnows appeared adversely affected up to 0.6 mile downstream in two of the four plants. Total residual chlorine concentrations less than 0.1 mg/l were toxic to fathead minnows. Rainbow trout 96-hour total residual chlorine TL-50 concentration below two plants was 0.023 mg/l. (Jones-Wisconsin)

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Chlorinated Municipal Waste Toxicities to Rainbow Trout and Fathead Minnows

by

Bureau of Water Management
Michigan Department of Natural Resources
Lansing, Michigan 48926

for the

ENVIRONMENTAL PROTECTION AGENCY

Grant Number 18050 GZZ
October 1971

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ABSTRACT

This project consisted of separate studies at four different Michigan municipal wastewater treatment plants. Ten rainbow trout (Salmo gairdneri) and ten fathead minnows (Pimephales promelas), each previously acclimated to the river, were held for 96 hours in live boxes in the receiving stream above and below these plant outfalls. Fish held below these outfalls were subjected to both chlorinated and non-chlorinated exposures during effluent discharge. During fish exposure, the test waters were monitored chemically and bacteriologically.

Total residual chlorine concentrations below three of the four plants were toxic to rainbow trout at distances up to 0.8 mile. Fathead minnows appeared adversely affected up to 0.6 mile downstream in two of four plants. Total residual chlorine concentrations less than 0.1 mg/l were toxic to fathead minnows in the plants.

The rainbow trout 96-hour total residual chlorine TL-50 concentration below two plants was 0.023 mg/l.

This report was submitted in fulfillment of Project Number 18050 GZZ under the (partial) sponsorship of the Water Quality Office, Environmental Protection Agency.

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SECTION I

CONCLUSIONS

1. Total residual chlorine concentrations below three of four Michigan municipal wastewater treatment plants were toxic to rainbow trout after four or more days. Fathead minnows were less affected by the chlorinated wastes with toxicity observed below two of the four plants.

2. Long river reaches were rendered uninhabitable to many fish due to the detrimental effect of chlorinated Michigan municipal effluents. Stream reaches up to 0.8 mile long below these municipal outfalls were lethal to rainbow trout after four days. Fathead minnows were adversely affected up to 0.6 mile below two of four plants.

3. The major factor affecting the downstream extent of the toxicity to rainbow trout was the waste dilution by the river. In two instances after the waste was thoroughly mixed the waste was toxic up to 0.7 mile farther downstream.

4. Rainbow trout 96-hour TL-50's in two rivers were 0.014 and 0.029 mg/l. Results pooled from these two plants showed the total residual chlorine 96-hour TL-50 of rainbow trout to be 0.023 mg/l. Lethal levels for fathead minnows were less than 0.1 mg/l.

SECTION II

RECOMMENDATIONS

With the large number of municipal and industrial plants that discharge chlorinated wastes, the results of this study and others, indicate the need to further document the effect of chlorinated wastes on fish life and other aquatic life in the receiving streams.

This project was conducted during the winter months when fish survival and river conditions should be most favorable. A parallel study during the summer months should be conducted to determine the toxicity of these wastes under less favorable conditions.

An effort should be made to assess the affect of municipal discharges on the upstream migration of anadromous fishes. This information would be valuable in Michigan because of the extensive introduction of anadromous fishes.

Public health considerations require adequate disinfection of wastes but present chlorination practices can be toxic to aquatic life. Until ecologically proven safe disinfection practices are developed, it is recommended that in-plant monitoring of chlorine be carefully controlled and a more accurate analytical method be found to measure chlorine other than the orthotolidine color comparator.

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SECTION III

INTRODUCTION

Recent research at the Duluth National Water Quality Laboratory and the Michigan Water Resources Commission have indicated an urgent need for further work on the effect of chlorinated municipal wastes on the aquatic life in receiving streams.

This project attempted, under field conditions, to assess the effect of these wastes on the fish life and to help support or reject laboratory findings of other researchers. Further objectives were to arrive at tolerance levels and determine the length of river below each plant outfall rendered unavailable to resident fish populations.

The extreme toxicity of free chlorine to fish has long been recognized. Doudoroff and Katz (1950) cite early studies by Taylor and James (1928) and Wilhelmi (1922). A number of more recent papers are listed by McKee and Wolf (1963). In one of the most recent efforts Dandy (1967), working with brook trout and investigating the responses of this species when exposed to test chemicals, found a lethal threshold for a seven day exposure of 0.01 parts per million (ppm) free chlorine.

Chlorine present in municipal wastewater discharges is alsmost always present in the combined form. It is usually combined with ammonia, ammonium hydroxide or ammonium ions to form mono-, di-, or trichloramine. Sawyer and McCarty (1967) also list organic and inorganic reducing agents, phenols, and organic compounds with unsaturated linkages as substances which will react with free chlorine. Coventry, Shelford and Miller (1935) found that water with a 0.05 ppm chloramine content killed trout fry within 48 hours. A recent author, Zillich (1970), has shown that chloramines associated with chlorinated municipal sewage had a 96-hour 50 percent tolerance level (TL-50) in the range of 0.05-0.16 mg/l. Arthur and Eaton (1971) found that the lowest concentrations having no detrimental effects to fathead minnows (Pimephales promelas) and amphipods (Gammarus pseudolimnaeus) were 16 and less than 3.4 micrograms per liter (ug/l), respectively.

Various researchers have investigated the comparative toxicities of free chlorine and chloramines. Merkens (1958), using a continuous-flow bioassay set-up with rainbow trout as test fish, found the toxicity of free chlorine and the chloramines all of the same order with free chlorine being the most toxic. He found a safe concentration in the range of 0.004-0.08 ppm total residual chlorine. McKee and Wolf (1963) cite Westfall who found chloramines more toxic than free chlorine to warmwater fish; Westfall also reports 0.06 mg/l chloramines being lethal to trout fry. McKee and Wolf also cite work done by the Washington Department of Fisheries using salmon as test fish. They found, in aerated freshwater, chloramines were more toxic than free chlorine.

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Field investigations of the effects of chlorine have been noticeably lacking. One author (Tsai, 1968 and 1970) conducted field surveys of the fish fauna in three Maryland streams receiving chlorinated wastes. He found that the number of species and fish abundance decreased drastically in the area immediately below chlorinated sewage outfalls. Downstream from these outfalls, in organically enriched areas, the fish community composition changed although there were no changes in species diversity indices calculated. He also reported that during their spawning season upstream migrations of white catfish and white perch were blocked by the chlorinated sewage effluents concentrated in the area immediately below the sewage outfalls.

Wuerthele (1970 a and b) held fathead minnows in live boxes above and below a Michigan wastewater treatment plant outfall. He found complete mortality four miles downstream. Interpretation of results from this study was complicated by the presence of industrial wastes in the municipal waste and an industrial discharge upstream from the municipal outfall.

SECTION IV

METHODS

Suitable study locations for this project were selected with the cooperation of the basin engineers in the Wastewater Section of the Michigan Department of Public Health. The final selection of wastewater treatment plants (WWTP) discharging to Michigan rivers was based on the following criteria:

a. Absence of any public health danger when chlorination was temporarily discontinued.

b. Absence of any other toxicants in the river or coming into the WWTP.

c. Plant's effluent comprising a significant portion of the river flow.

d. Presence of dependable operators at the plant.

e. Reasonably accessible conditions.

An effort was made to get a geographical distribution of plans as well as a representative sampling of various types of waste treatment. The total project consists of studies at four different WWTP's. Each study included a chlorinated phase and a non-chlorinated phase, during which separate sets of fish were exposed to chlorinated and non-chlorinated wastes. The general procedure at each study area was as follows: Fish were held in cages in the river above and below an outfall. The fish held below the outfall were subjected to chlorinated wastes for a period and mortalities noted. The treatment plant stopped chlorinated, new fish were introduced and exposed for a similar period to non-chlorinated wastes. While the fish were exposed, chemical and bacteriological samples were taken to monitor possible causes of mortality other than chlorine. Since sites were chosen where there were no other known toxic materials coming into the plant or present in the river, any differences in survival between the two exposures could be assumed to be due to the chlorinated compounds in the waste.

In the preliminary portion of each study Rhodamine B dye was used to determine the location of the WWTP's discharge plume. Stations were located in this plume and samples taken for total residual chlorine determination. Station locations were chosen so as to have a wide range of total residual chlorine concentrations. At each station, two seven foot steel fence posts were driven into the river bottom approximately 15 inches apart. One test cage was suspended from each post. The test cages, constructed of 3/4 inch exterior plywood, each held a volume of nearly one cubic foot. Vertical window screen openings were provided to allow limited water circulation

(Figure 1). The test cages were suspended on the posts with U-bolts and the two cages wired together to prevent oscillation in the river current. Suspending the cages by the U-bolt arrangement allowed vertical movement with changes in river stage. When the cages were in place, approximately 2 1/2 inches of each cage was above the water surface. At each upstream control station plywood stock cages were placed to hold the stock of fish. The stock cages were constructed of 3/4 inch exterior plywood and held a volume of nearly six cubic feet.

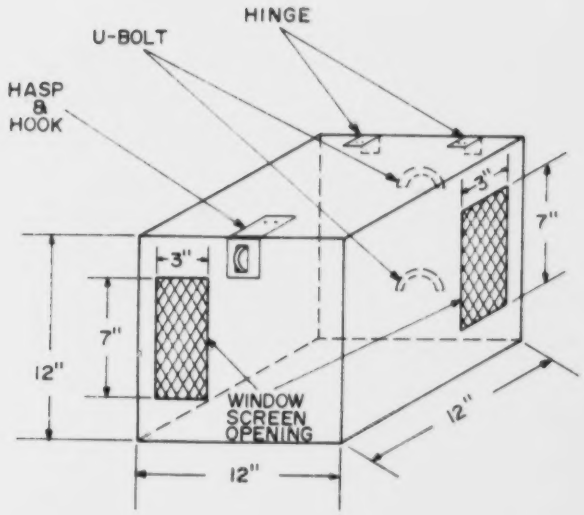


Figure 1. Test cage construction and dimensions.

Window screen openings were provided to allow limited water circulation (Figure 2). These cages were also held in the river by two seven foot steel fence posts placed through U-bolts on both ends of each cage. A staff gage was also placed at the upstream control station and was read each time a sample was taken.

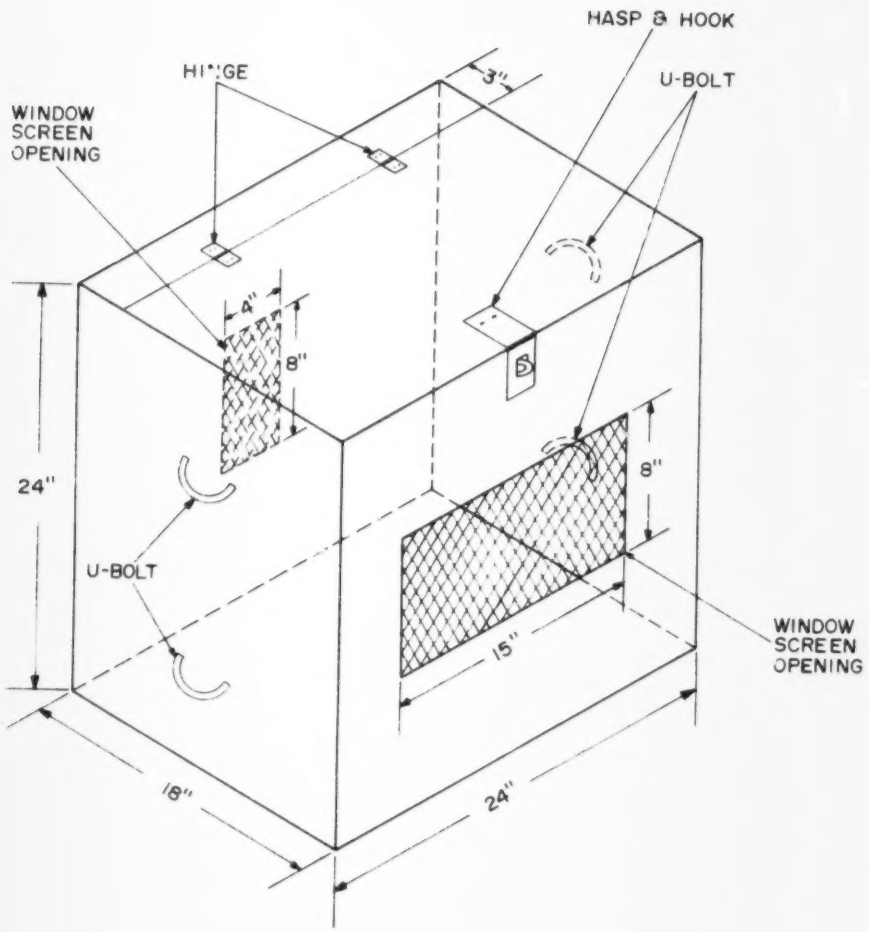


Figure 2. Stock cage construction and dimension.

Two species of test fish, rainbow trout (Salmo gairdneri) and fathead minnows (Pimephales promelas) were obtained from the Michigan Department of Natural Resources Wolf Lake Fish Hatchery and hauled to the study sites in a 144-gallon truck mounted water tank. The minnows and trout ranged in size from 2-3 inches and 4-5 inches, respectively. For six days prior to each phase, 500 fish of each species were placed in separate stock cages and acclimated to the river. At the start of each phase, the fish were transported from the stock cages to the downstream test cages in a plexiglass aquarium. Ten fish of each species were rapidly and carefully placed in each test cage.

Samples were collected at each station and from the final effluent once a day, usually at noon, throughout each phase. These samples were analyzed for total residual chlorine, dissolved oxygen (DO), pH, temperature, and total and fecal coliform organisms. In addition, ammonia (NH_3) samples were collected from the effluent and the upstream control station. Once during each phase, samples were collected every four hours at each station starting at 8:00 a.m. for a 24-hour period. During these diurnal sample collections DO, pH, temperature and chlorine determinations were made. During the noon and 4:00 a.m. collections, NH_3 and bacterial samples were also taken. Noon and 4:00 a.m. were normally the times at which the largest and smallest volumes of waste were treated. Samples were also taken from the river at the upstream control station and from the WWTP final effluent and analyzed for various possible industrial toxicants.

Dissolved oxygen, pH, temperature, and chlorine determinations were made by Michigan Water Resources Commission (MWRC) field personnel in each WWTP laboratory while temperature determinations were made in the field. Dissolved oxygen determinations were made by the azide modification of the Winkler Method as given in Standard Methods. Determination of pH was made with a glass electrode pH meter manufactured by the Beckman Instrument Company. Total residual chlorine determinations were performed using a Fisher and Porter Model 17T1010 amperometric titrator. Ammonia and all other chemical determinations were performed at the Michigan WRC Wastewater Laboratory in Lansing using methods described in Standard Methods. Bacteriological samples were analyzed using the membrane filter technique by Michigan Department of Public Health personnel in Lansing.

Michigan WRC Hydrological Survey personnel gaged each stream to determine river flows. In each instance the river was gaged at the upstream control station and values found used as the basis for later computation of river flows. River flows were estimated by two methods, 1) from United States Geological Survey (USGS) rating curves if available or 2) by calculations. Flows were calculated assuming that the river had vertical banks and that for the changes in river stage observed there was no change in velocity. For any change in stage the corresponding change in cross sectional area was calculated. This cross sectional area was added to the cross sectional area at the time the river was gaged. The new cross sectional area was multiplied by the original velocity giving the new river flow. This is at best an extremely rough estimate of the river flow, however, it was the only estimate of flow available.

Survival of each species was observed after 48 and 96 hours of exposure in each phase. Fish were counted as dead if there was no discernible muscular or opercular movement and no visible heartbeat. Initially a fish check was not performed after 48 hours because it was felt that lifting and opening the cages might unnecessarily stress the fish. The first scheduled non-chlorinated phase at Charlotte was washed out after 72 hours by a heavy rain and runoff from melting snow. In subsequent studies a 48-hour fish check was included. By checking fish after 48 hours in both phases it would be possible to salvage 48-hour survival data from any later washed out phases.

SECTION V

STUDY 1

Description: The Battle Creek River at Charlotte was the first study site. The reach studied was from US-27 in Charlotte to the Broadway Highway Bridge, a total distance of approximately two river miles (Figure 3). The river flows through low farm land and partially wooded pasture. The river width varied from 15 to 60 feet and the depth ranged from 1 to 4 feet. The bottom is generally sand and mud except for isolated gravel and sand reaches. Calculated river flows varied from 21 to 89 cfs. Like many southern Michigan warmwater streams, this river contains suckers, smallmouth bass, pike, dog fish and carp.

Waste treatment at the Charlotte WWTP, which serves 8,200 people, consists of primary settling, trickling filter secondary treatment, final settling and gas chlorination. Mean daily plant discharges for February and March were 0.8955 and 0.7512 million gallons per day (mgd), respectively (Appendix 2)

Results and Discussion: The fish were held for 120 hours in each phase rather than 96 hours. Subsequent studies were conducted for 96 hours.

During the chlorinated phase, the plant operators attempted to maintain a chlorine residual of a trace to 0.5 mg/l. To maintain this level, the operators at the end of the workday (8:00 p.m.), decreased the chlorinator settling to 1/2 of the daytime setting. Residuals measured by plant personnel with the orthotolidine arsenite color comparator ranged from 0.0 to 2.0 mg/l and were consistently lower than amperometric titrator determinations. Concurrent amperometric titrator total residual chlorine concentrations ranged from 0.96 to 2.94 mg/l and averaged 1.77 mg/l (Appendix 1). Before the fish were placed in the test cages for the non-chlorinated exposure (March 2-7) the chlorine was turned off for 24 hours to ensure that no residual chlorine remained.

Results in Table 1 show almost a complete kill of both species during the chlorinated phase 0.6 mile downstream. Station 1, the upstream control, and Stations 2 and 6, where no chlorine was detected, had low mortality. Survival at Stations 2 and 3 graphically demonstrates the extreme waste toxicity. These stations were approximately 10 feet apart, yet Station 3, in the discharge plume, had no survival, while Station 2 averaged 85 to 89 percent survival of trout and minnows, respectively. Survival of both species at Station 3 during the non-chlorinated phase was high. Results similar to Station 3 were also noted at Stations 4 and 5 during the respective phases.

The minnow survival data was highly variable during both phases. It is obvious that the chlorine compounds were toxic to minnows but due to this variance no statistical analyses were attempted.

Figure 3. Map of Charlotte area showing station locations.

Station	Location
1	Midstream, 80 yards upstream from outfall
2	Midstream, at outfall, not in the discharge plume
3	Right bank, 10' downstream from outfall directly in plume
4	Midstream, 150 yards downstream from outfall
5	Midstream, approx. 0.6 mile downstream, 75' upstream from Kalamo Rd.
6	Left bank, approx. 2 miles downstream from outfall, 50' downstream from Broadway Highway

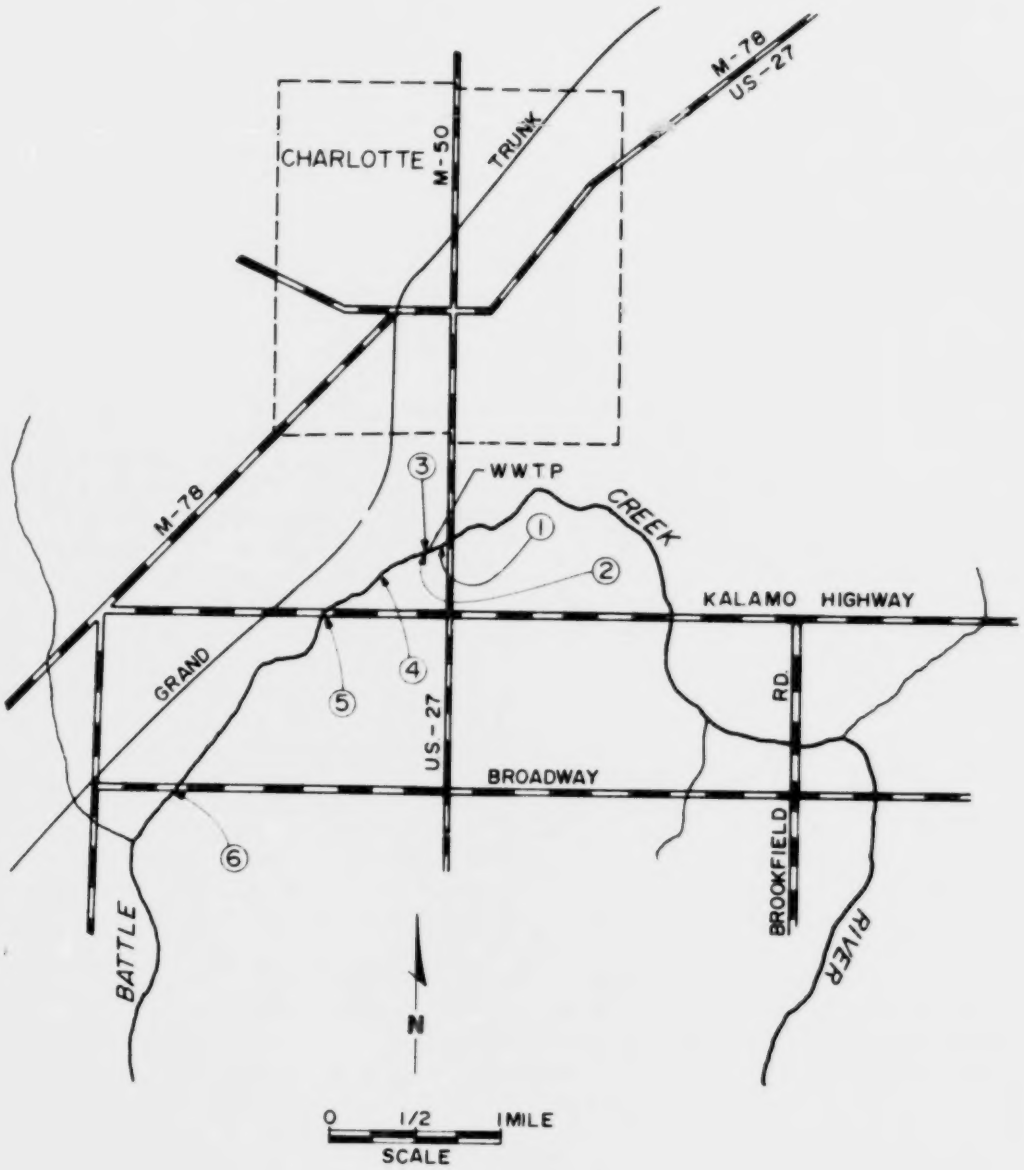


Table 1. Percent survival of fish after 120 hours below the Charlotte WWTP outfall.

SPECIES-PHASE		STATIONS 1	2	3	4	5	6
Rainbow Trout							
Chlorinated	R[1]	80	80	0	0	0	90
	L	70	90	0	0	10	90
Non-chlorinated	R	100	100	100	100	100	100
	L	100	100	100	100	100	100
Fathead Minnows							
Chlorinated	R	80	77[2]	0[2]	0[2]	0[2]	40
	L	90	100	0	0	25	90
Non-chlorinated	R	100	80	60	50	40	70
	L	50	70	20	90	50	90
Average Total Residual Chlorine Concentration (mg/l)[3]		0.000	0.000	0.647	0.045	0.007	0.000

(1) R = right cage, L = left cage

(2) More than 10 fathead minnows initially placed in test cages during chlorinated phase.

(3) Measured with an amperometric titrator.

Threshold lethal concentrations for both species would be in the range of 0.0 to 0.03 mg/l, the range of values at Station 5 where both partial survival and total residual chlorine were found.

Calculated mean daily river flows were higher in the non-chlorinated phase, 64.5 versus 28.7 cfs (Appendix 1). It could be argued that due to lower flows in the chlorinated phase increased concentrations of other waste materials caused the mortality. High survival at Station 3 during the non-chlorinated phase in nearly undiluted waste indicates that the mortality was due to the chlorinated compounds and not some other toxicant.

Other parameters (DO, pH, temperature and NH_3) did not differ appreciably between the two phases (Appendix 1). High ammonia concentrations were found in the effluent in both phases. High survival at all stations during the non-chlorinated phase indicated that the mortality was not due to the ammonia. Other possible toxicants were not found in high concentrations in either the river or effluent (Table 2).

Table 2. Characteristics of Charlotte WWTP effluent and river water.

Parameter[1]	River	Effluent
Time	4:00 p.m.	4:15 p.m.
Temperature	1°C	7.5°C
DO	11.0	10.0
COD	32	310
pH	7.8	7.8
NO_3-N	1.4	0.10
Chlorides	12	260
NH_3-N	0.31	27
NO_2-N	0.02	0.05
Alkalinity	195	405
Hardness	300	415
CN	0.00	0.00
CR_2+6	0.00	0.00
Pb	0.0	0.0
Ni	0.0	0.0
Cu	0.00	0.1
Cd	0.00	0.00
Zn	0.00	0.10

[1]All parameters except pH expressed as mg/l.

Conclusion: Total residual chlorine concentrations in the Charlotte WWTP effluent was extremely toxic to both fish species. A river reach of 0.6 mile below the outfall could not support rainbow trout or fathead minnows for 5 days. Extremely low total residual chlorine concentrations (0.0 to 0.03 mg/l) were found toxic to both species.

SECTION VI

STUDY 2

Description: Ogemaw Creek at West Branch in Ogemaw County was the second study site. The river reach studied was from approximately 100 feet upstream from the West Branch WWTP discharge to approximately 50 yards downstream from the Flowage Lake dam, a total distance of approximately 1.8 river miles (Figure 4). The watershed consists mainly of second growth forest wood lots and isolated farms. Trees and overhanging brush are the predominant river bank cover. The river bottom is sand, silt and gravel. Because of a steep gradient, few pool areas were found except immediately above Flowage Lake. Widths varied from 15-40 feet and depth from 1-3 feet. Calculated river flows varied from 14 to 45 cfs. Fish commonly found are brown and brook trout, northern pike, perch, largemouth bass, suckers, bullheads and various minnows.

The West Branch WWTP serves approximately 2,000 people with treatment consisting of primary settling and gas chlorination. Daily volumes discharged during March varied from 220,000 to 321,000 gallons per day and averaged 246,000 gallons.

Results and Discussion: Prior to the non-chlorinated phase (March 8-12, 1971) the chlorination was shut off for three days. In the chlorinated phase (March 15-19, 1971) the plant operator tried to maintain a chlorine residual of 0.5 mg/l. To maintain this residual, the chlorine residual was checked and chlorinator adjustments made every two hours throughout the 96 hours. The range in chlorine residual observed by the operators using a color comparator was 0.0-1.2 mg/l. The range found with the amperometric titrator over the same period was 0.95-1.89 mg/l and averaged 1.35 mg/l.

Results in Table 3 show that only 2 of 20 trout survived for 96 hours at Station 3 and 10 of 20 at Station 5 (Table 3). Station 4, in Brewery Creek, was included to insure the absence of any toxicants coming from this tributary. Survival of both species at Station 4 during both phases of the study was excellent. Trout mortality at Stations 2, 3 and 4 did not occur until the second half of the chlorinated exposure (Table 4). It is difficult to determine if this mortality is due to a cumulative exposure effect or to higher total residual chlorine concentrations in the second half of the chlorinated exposure since river flows steadily decreased, being 20 cfs lower at the end of the chlorinated exposure than they were at the beginning (Appendix 3).

Figure 4. Map of West Branch area showing station locations.

Station	Location
1	Right bank, 100' upstream from outfall
2	Midstream, 125' downstream from outfall
3	Midstream, 300' downstream from outfall
4	Midstream, Nelson Creek, 40' up from confluence with Rifle River
5	Midstream, 150' downstream from confluence with Nelson Creek, 500' below outfall
6	Midstream, 0.6 mile downstream from outfall at M-76 bridge
7	Midstream, 1.3 miles downstream at inlet to Flowage Lake
8	Midstream, 100 yards downstream from dam, approximately 1.8 miles downstream from outfall

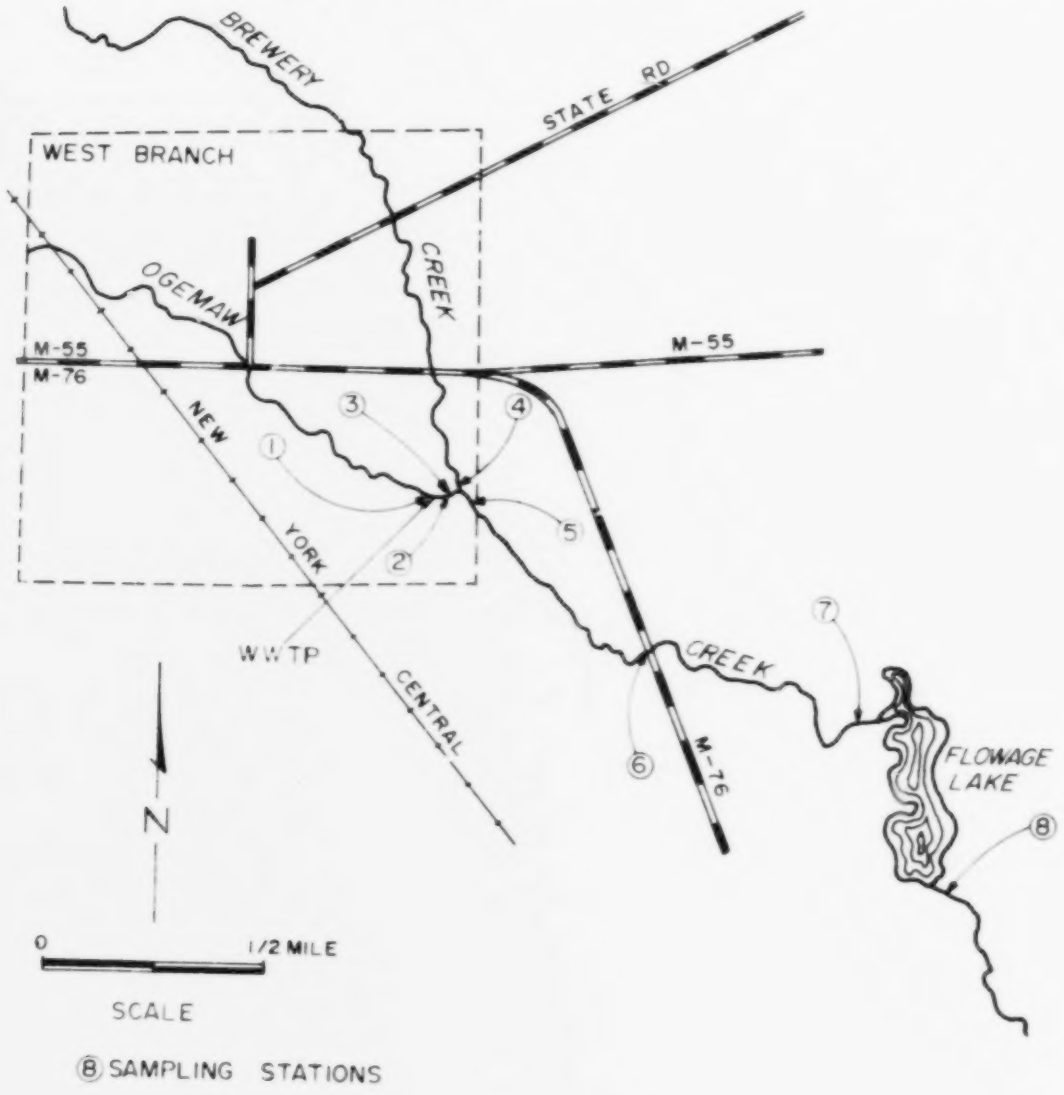


Table 3. Percent suvival of fish after 96 hours below the West Branch WWTP outfall.

SPECIES-PHASE		STATIONS 1	2	3	4(2)	5	6	7	8
Rainbow Trout									
Chlorinated	R(1)	90	0	0	-	50	100	100	100
	L	100	0	20	100	50	100	100	100
Non-chlorinated	R	100	100	100	-	100	100	100	100
	L	100	100	100	100	100	90	100	100
Fathead Minnow									
Chlorinated	R	70	90	80	-	80	70	90	80
	L	70	60	70	80	60	90	90	90
Non-chlorinated	R	70	90	90	-	70	90	90	70
	L	90	100	40	100	100	100	90	90
Average Total Chlorine Concentration (mg/l)(3)		0.000	0.018	0.032	0.000	0.014	0.002	0.000	0.000

(1) R = right cage; L = left cage

(2) Station 4 in feeder stream, only one cage used

(3) Measured with an amperometric titrator

* * *

Table 4. Percent survival of fish after 48 hours of chlorinated exposure below the West Branch WWTP outfall.

Station	Rainbow Trout Right	Rainbow Trout Left	Fathead Minnow Right	Fathead Minnow Left
1.	90	100	80	70
2.	90	100	100	100
3.	80	100	100	90
4(1)	100		100	
5.	90	80	100	90
6.	100	100	100	90
7.	100	100	100	100
8.	100	100	90	90

(1) Station 4 in feeder stream, only one cage used.

At Stations 5 and 6, total residual chlorine was only detected during the second half of the exposure indicating that the mortality in Station 5 occurred due to the increased total residual chlorine concentrations during the second half.

Interpretation of the minnow survival data is complicated by the mortality in the controls during both phases. Complete minnow mortality was not observed at any station suggesting that total residual chlorine was not high enough to kill this species in this river during a 96-hour exposure.

Dose-response data typically yields a sigmoid type curve. It was assumed for the purpose of this report that the inflection region of the dose-response curve was linear. Linear regression was then employed to determine both the dose-response relationship in this inflection region and 96-hour TL-50's. In calculating the regression the lowest concentration at which no fish survived and the highest concentration at which all fish survived were assumed to encompass this inflection region. Data outside this region was not included in the regression calculation. The regression was calculated for log of mean total residual chlorine concentration on percent survival at three stations in this test.

The r value was significant beyond the 0.01 level (Steel and Torrie, 1960), implying that this relationship did account for a significant portion of the variance in the inflection region of this dose-response data. The 96-hour TL-50 concentration taken from this regression line is approximately 0.014 mg/l (Figure 5).

Calculated mean daily river flows during the non-chlorinated phase were lower than during the chlorinated phase, 17 versus 29 cfs (Appendix 3). Decreased dilution of the non-chlorinated waste and excellent survival of trout is added proof that the high mortality in the chlorinated phase is due to the chlorine compounds. Unlike Charlotte, West Branch results are not confounded by high waste ammonia concentrations. Mean ammonia concentrations in the effluent in both phases at West Branch were 1.88 and 3.26 mg/l. High concentrations of other toxicants were not found in either the river or the effluent (Table 5) and DO, temperature and pH did not differ appreciably in the two phases (Appendix 3).

Conclusion: Total residual chlorine concentrations below the West Branch WWTP outfall were toxic to rainbow trout but not to fathead minnows. Five out of ten rainbow trout in each cage died after 96-hours approximately 500 feet downstream. The average total residual chlorine concentration at this station was 0.014 mg/l. The rainbow trout 96-hour TL-50 concentration obtained was 0.014 mg/l.

Figure 5. Chlorine concentration and regression for percent survival of rainbow trout below the West Branch WWTP outfall.

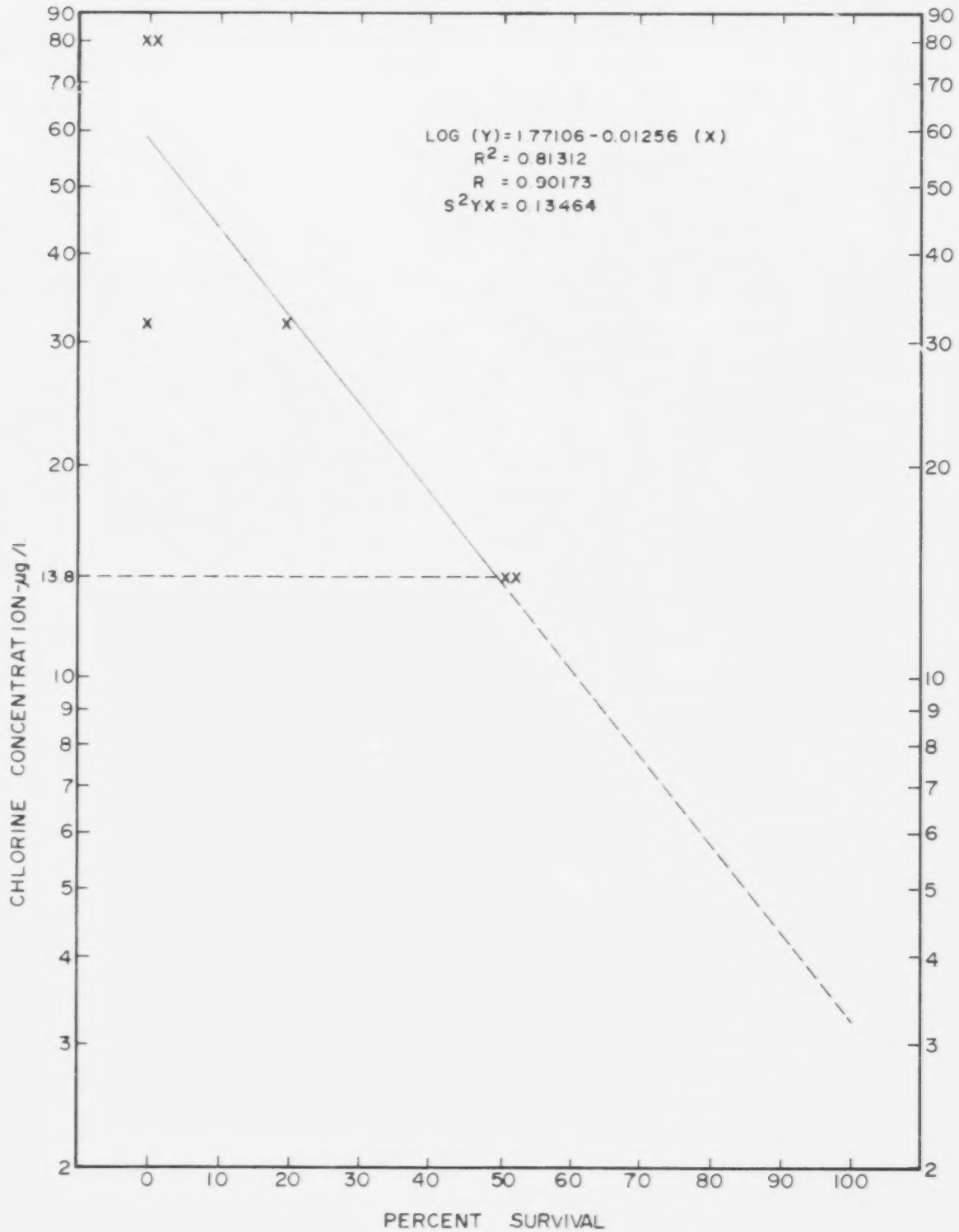


Table 5. Characteristics of West Branch WWTP effluent and river water.

Parameter[1]	River	Effluent
Time	12:00 p.m.	3:45 p.m.
Temperature	1°C	8°C
COD	11	110
Total Solids	242	480
pH	8.2	7.6
NO_3-N	0.20	0.30
Chlorides	27	97
NH_3-N	0.24	2.4
Conductivity	390	710
NO_2-N	0.01	0.03
Alkalinity	155	210
Hardness	180	240
CN	0.00	0.00
Cr^{+6}	0.00	0.00
Pb	0.0	0.0
Ni	0.0	0.0
Cu	0.0	0.0
Cd	0.00	0.00
Zn	0.00	0.05

(1) All parameters except pH and conductivity expressed as mg/l.

SECTION VII

STUDY 3

Description: The South Branch of the AuSable River at Roscommon, Roscommon County, was the third study area. The reach studied was from approximately 50 yards upstream from the Roscommon WWTP outfall to a point approximately 0.5 mile downstream from the Roscommon WWTP discharge, a total distance of approximately 0.6 mile (Figure 6). The watershed is almost entirely timbered. The bank cover consists mainly of tag alders, cedars, and mixed hardwoods. The stream bottom is mainly sand and small gravel with isolated areas of mud along the banks. The width varied from 50 to 80 feet averaging approximately 60 feet. Depths at all stations varied from 3-5 feet. The calculated river flows during this test varied from 119 cfs to 179 cfs. Large numbers of brown trout were introduced in the AuSable in the early 1920's. They remain the dominant sport fish, maintaining excellent populations of wild fish through natural reproduction. Brook trout are commonly found in the diverse fish population which also includes various minnows, sculpins, darters, small and largemouth bass, redhorse, suckers, pike and members of the sunfish family.

Treatment at Roscommon's plant, which serves 900 people, consists of Imhoff tank primary settling followed by gas chlorination. The mean daily volume discharged during March 1971 was 47,890 gallons.

Results and Discussion: During the chlorinated phase (March 8-12, 1971) the operator maintained a chlorine residual concentration of 2-2.5 mg/l as measured by the orthotolidine color comparator method. Effluent total residual chlorine concentrations measured by amperometric titration ranged from 5.01 to 32.5 mg/l, averaging 18.92 mg/l. The range of values in the color comparator was 0.0 to 2.5 mg/l which accounts for the failure to detect higher chlorine residuals. The chlorination was turned off three days prior to the non-chlorinated phase (March 15-19, 1971) to allow all residual chlorine to dissipate.

Chlorine was only detected twice during this study, once each at Stations 2 and 4 (Appendix 5). The average volume of waste discharged was 50,600 gallongs (0.078 cfs) (Appendix 6). This volume comprised only 0.06 percent of the river. This large a dilution would account for the failure to detect chlorine in the river when such high effluent residuals are found.

No excessive mortality was observed in trout or minnows except at Station 3, 50 yards below the Roscommon WWTP discharge (Table 6). Comparison of the 48 and 96-hour results show that this mortality occurred between 48 and 96 hours (Table 7).

Figure 6. Map of Roscommon area showing station locations.

Station	Location
1	Midstream, 50 yards upstream from outfall
2	Midstream, 20 feet downstream from outfall
3	Left bank, 50 yards downstream from outfall
4	Midstream, 100 yards downstream from outfall
5	Left bank, 1/2 mile downstream from outfall

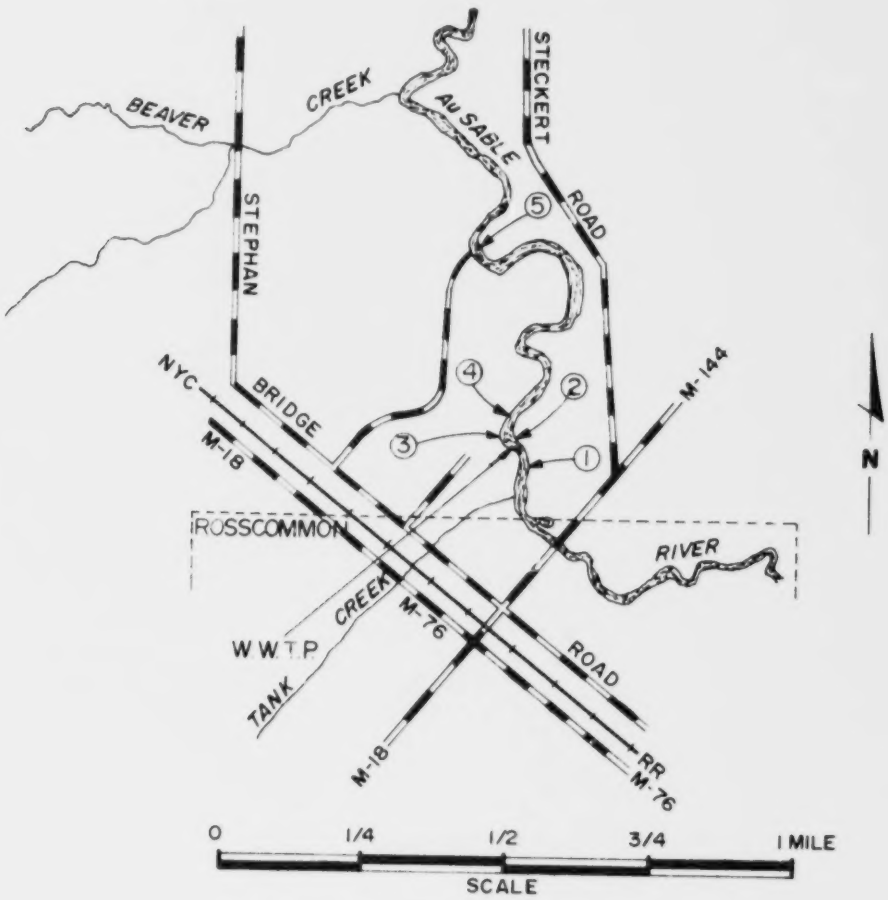


Table 6. Percent survival of fish after 96 hours below the Roscommon WWTP outfall.

SPECIES-PHASE		STATION 1	2	3	4	5
Rainbow Trout						
Chlorinated	R[1]	90	100	20	100	100
	L	100	100	0	100	100
Non-chlorinated	R	100	100	100	100	100
	L	100	100	100	100	100
Fathead Minnow						
Chlorinated	R	100	80	80	100	100
	L	100	80	90	100	80
Non-chlorinated	R	80	100	80	90	80
	L	100	50	80	90	80
Average Total Residual Chlorine Concentration (mg/l)[2]		0.000	0.001	0.000	0.002	0.000

[1] R = right cage; L = left cage

[2] Measured with an amperometric titrator

* * *

Table 7. Percent survival of fish after 48 hours of chlorinated exposure below the Roscommon WWTP outfall.

Station	Rainbow Trout Right	Rainbow Trout Left	Fathead Minnow Right	Fathead Minnow Left
1	100	100	100	100
2	100	100	100	100
3	100	100	100	90
4	100	100	100	100
5	100	100	100	100

No mortality was observed at Station 2, 20 feet downstream from the outfall. From field observations it appears that the river stage went down and the plume moved to the center of the river away from Station 2. Stations 3 and 4 were in the plume but the high dilution could account for failure to detect chlorine.

Other possible toxicants were not found in high concentration in either the river or the effluent (Table 8). Temperature, DO, pH and ammonia did not differ appreciably between the two phases at any station (Appendix 5).

Table 8. Characteristics of Roscommon WWTP effluent and river water.

Parameter[1]	River	Effluent
Time	10:30 a.m.	10:30 a.m.
COD	22	150
Total Solids	100	444
pH	7.5	7.3
NO_3-N	0.2	0.7
Chlorides	7	140
NH_3-N	0.03	12
Conductivity	210	850
NO_2-N	0.00	0.06
Alkalinity	95	205
Hardness	110	220
CN	0.00	0.00
Cr^{+6}	0.00	0.00
Pb	0.0	0.0
Ni	0.0	0.0
Cu	0.0	0.0
Cd	0.00	0.00
Zn	0.00	0.05

[1]All parameters except pH and conductivity expressed as mg/l.

Conclusion: High dilutions and changing river stage resulted in only two instances of total residual chlorine detection. Trout mortality was observed at only one station. Since total residual chlorine was not detected at this station it is impossible to attribute the mortality to chlorine compounds present in the waste.

SECTION VIII

STUDY 4

Description: Sycamore Creek below Mason, in Ingham County, was the fourth study site. The reach studied was from 50 yards upstream from the Mason WWTP outfall downstream to the US-127 bridge, a total distance of approximately four river miles (Figure 7). Land use in the watershed is mainly agricultural. The creek's width varied from 15 to 40 feet and the depth varied from 1 to 3 feet. The bottom is sand and mud downstream from the Mason WWTP outfall, while upstream some gravel riffles were observed. The calculated river flows varied from 21 to 25 cfs. Species of fish found include large and smallmouth bass, pike, and various members of the sunfish and minnow families.

Treatment at the Mason plant, which serves 4,500 people, consists of primary settling, activated sludge secondary treatment, final settling and gas chlorination. Mean daily volumes discharged for March and April were 0.867 and 0.764 mgd, respectively (Appendix 8).

Results and Discussion: During the chlorinated phase (March 29-April 2, 1971) the operators attempted to maintain a chlorine residual of 1.5 mg/l but no chlorinator adjustment was made at the end of the work shift (4:00 p.m.). The residual chlorine concentrations observed by the Mason WWTP operator using a color comparator varied from 0.7 to 2.0 mg/l. Values found using the amperometric titrator varied from 1.82 to 3.89 and averaged 2.64 (Appendix 7). Prior to the non-chlorinated phase (April 5-9, 1971) the chlorinator was turned off for approximately 20 hours to ensure the absence of any chlorine residual.

Results in Table 9 show that at Station 5, 0.8 mile downstream, 7 out of 20 trout survived, and minnow survival was similar to the stations downstream where no total residual chlorine was observed. At Station 6, 1.5 miles downstream, there was partial mortality of both species probably due to natural mortality. Thus the chlorinated compounds in the Mason WWTP waste were toxic to trout at least 0.8 mile downstream and to minnows at least 250 yards downstream. Station 3, in Rayner Creek, was utilized to assure the absence of toxicants from this tributary. Survival of both species during both phases was excellent at this station. The mortality at Station 2 occurred in the first half of the exposure in both species (Table 10). At Station 4 mortality was observed in both halves of the exposure and at Station 5 trout died during the second half of the exposure.

Figure 7. Map of Mason area showing station locations.

Station	Location
1	Midstream, 100 feet upstream from outfall
2	Right bank 7 feet down from outfall in discharge plume
3	Midstream, Rayner Creek, 15 feet upstream, 175 yards downs from outfall
4	Right bank, 250 yards downstream from outfall
5	Midstream, 0.8 mile downstream from outfall at Howell Rd.
6	Midstream, 1.5 miles downstream from outfall
7	Midstream, two and one-half miles downstream from outfall at Harper Rd.
8	Midstream, four miles downstream from outfall - 40 yds. upstream from US-127 highway.

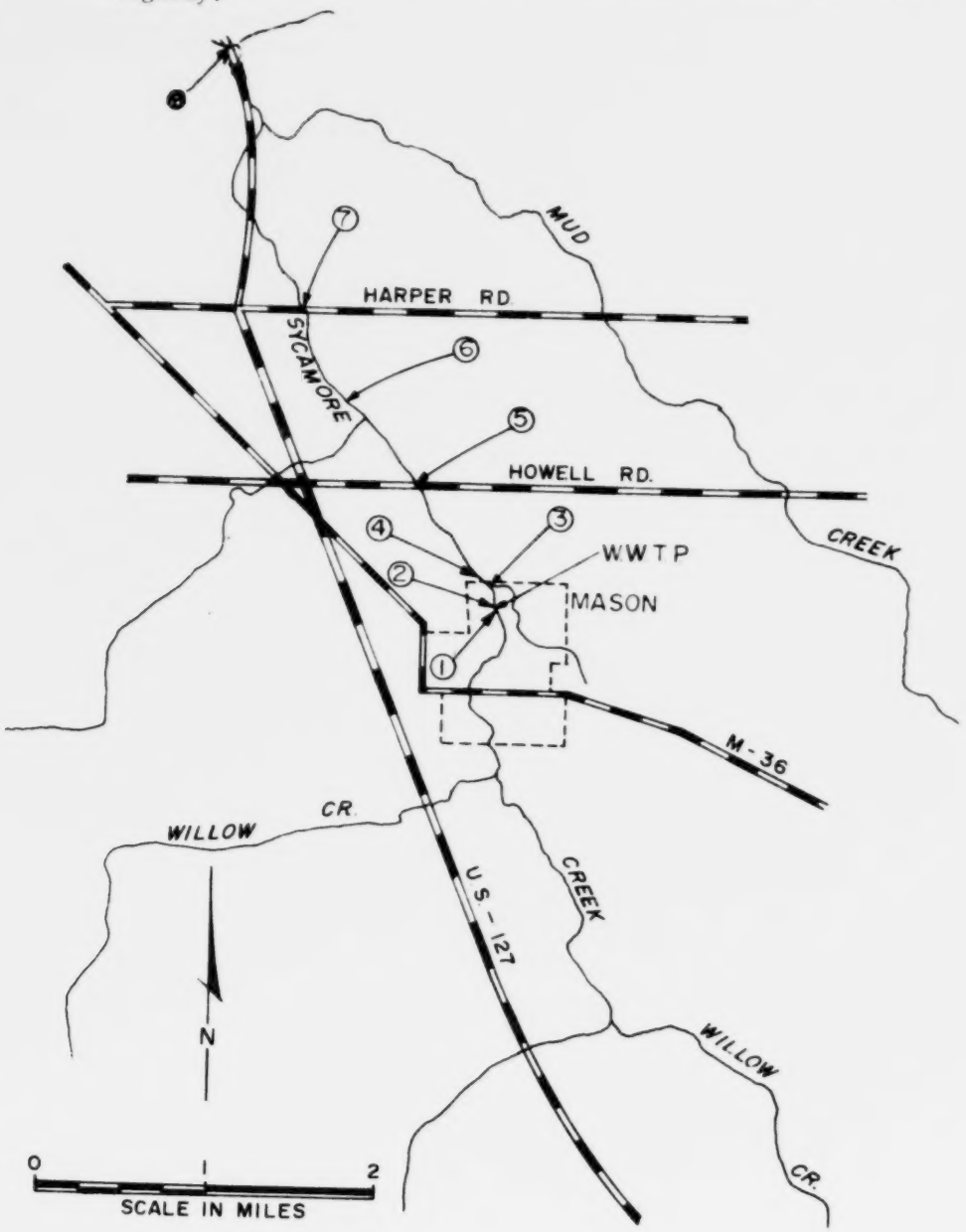


Table 9. Percent survival of fish after 96 hours below the Mason WWTP outfall.

SPECIES-PHASE		STATION 1	2	3[2]	4	5	6	7	8
Rainbow Trout									
Chlorinated	R[1]	100	0	100	0	30	100	100	100
	L	100	0		0	40	80	100	100
Non-chlorinated	R	100	100	100	100	(3)	100	100	100
	L	100	100		100	(3)	100	100	100
Fathead Minnow									
Chlorinated	R	70	0	90	30	60	100	90	70
	L	100	0		30	90	70	60	80
Non-chlorinated	R	90	100	100	100	(3)	80	90	90
	L	90	90		100	(3)	80	90	100
Average total residual chlorine concentration (mg/l)[4]		0.000	1.132	0.000	0.072	0.046	0.013	0.000	0.000

(1) R = right cage; L = left cage

(2) Station 3 is in feeder stream, only one cage used

(3) Both cages at this station were found missing after 24 hours

(4) Measured with an amperometric titrator

* * *

Table 10. Percent survival of fish after 48 hours of chlorinated exposure below the Mason WWTP outfall.

Station	Rainbow Trout Right	Rainbow Trout Left	Fathead Minnows Right	Fathead Minnows Left
1	100	100	100	100
2	0	0	0	0
3[1]	100		90	
4	30	60	80	90
5	100	90	90	100
6	100	100	100	90
7	100	100	90	90
8	100	100	90	90

(1) Station 3 in feeder stream, only one cage used.

Following the same procedure as was used at West Branch, linear regression was employed to analyze the survival data and determine 96-hour TL-50's. The rainbow trout linear regression, based on results at three stations, resulted in an r of 0.96043, significant beyond the 0.01 level, and an r^2 of 0.92242. Approximately 92 percent of the variance in these data can be accounted for by this regression. The regression line presented graphically in Figure 8 yielded a 96-hour TL-50 concentration of approximately 0.029 mg/l. The minnow regression was not significant implying that the relationship between total residual chlorine concentration and percent survival did not account for a significant amount of the variation in the inflection region of these data. Effluent and river samples contained no unusually high concentrations of other toxicants (Table 11). There was no appreciable difference in temperature, DO, and pH between the two phases. Ammonia concentrations were moderately high in the effluent and at Station 2, directly in the outfall, during both phases. The high survival observed in the non-chlorinated phase indicates that the ammonia concentrations observed were not toxic.

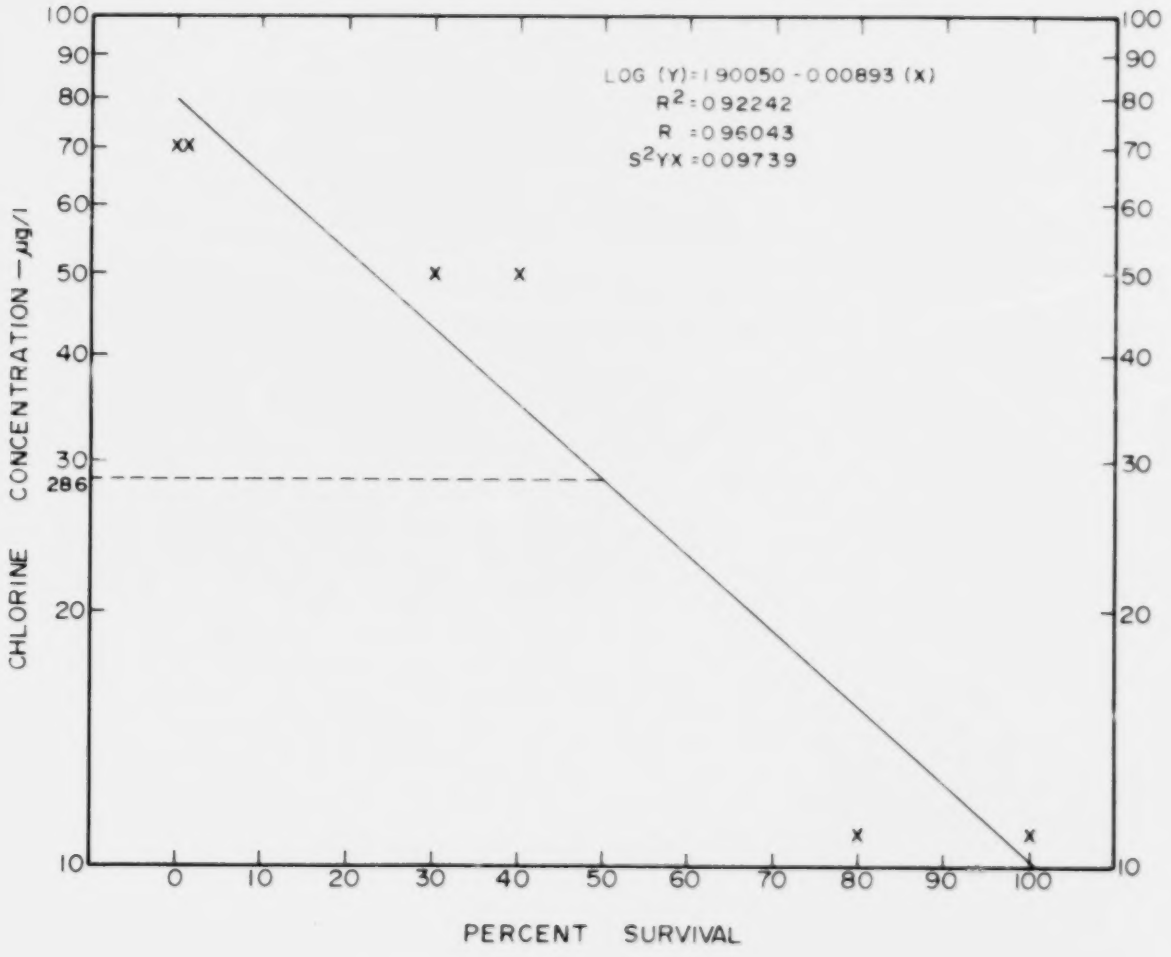
Table 11. Characteristics of Mason WWTP effluent and river water.

Parameter(1)	River	Effluent
Time	3:30 p.m.	3:30 p.m.
COD	17	150
pH	8.1	7.5
NO_3-N	2.7	0.34
NO_2-N	0.01	0.23
NH_3-N	0.02	13
Total PO_4	0.06	6.8
Conductivity	630	1,050
Alkalinity	215	380
Hardness	390	400
CN	0.00	0.00
Cr^{+6}	0.00	0.00
Ni	0.0	0.0
Pb	0.0	0.0
Cu	0.0	0.0
Cd	0.00	0.00
Zn	0.0	0.0

(1) All parameters except pH and conductivity expressed as mg/l.

Conclusion: Total residual chlorine concentrations below the Mason WWTP were highly toxic to both the rainbow trout and fathead minnows at a distance of 250 yards. For at least 0.8 mile downstream the waste was still toxic to rainbow trout but not to the fathead minnows. At this distance, the average total residual chlorine concentration observed was 0.05 mg/l. The 96-hour TL-50 found was 0.029 mg/l.

Figure 8. Chlorine concentrations and regression for percent survival of rainbow trout below the Mason WWTP outfall.



SECTION IX

GENERAL DISCUSSION

This project's primary objective was to determine if chlorinated WWTP discharges are actually lethal to fish present in the receiving streams. It is obvious from our results that these wastes were toxic to fish held downstream from these outfalls. In three of four plants the wastes were extremely toxic to rainbow trout. These wastes are not as lethal to fathead minnows since two of four tests showed toxicity. The downstream extent of the toxicity varied for rainbow trout from 500-4,200 feet and for fathead minnows from 750-3,200 feet (Table 12). In a similar experimental design Wuerthele (1970 a and b) found toxic reaches four miles long with fathead minnows but his results were complicated by the presence of other toxicants in his system.

The most important factor affecting the downstream extent of chlorinated municipal waste toxicity to rainbow trout was the waste dilution by the river. At Roscommon no mortality occurred from the chlorinated wastes with the high chlorinated residual maintained (18.92 mg/l) (Table 12). The volume of waste was extremely small and comprised only 0.06 percent of the river flow. At Mason the waste contained an average total residual chlorine concentration of 2.64 mg/l but comprised 5 percent of the river volume. Mortality was observed in rainbow trout 4,200 feet downstream from the Mason WWTP outfall.

It is important to note that even after the waste is thoroughly mixed rainbow trout mortality is still observable for considerable distances below the Mason and Charlotte plants (Table 12).

One author, Tsai 1968 and 1970, has shown that chlorinated WWTP wastes blocked the spawning runs of two species of estuarine semi-anadromous fishes. Similar conditions are feasible below Michigan WWTP's when the wastes are thoroughly mixed and still toxic to the anadromous rainbow trout.

Actual lethal concentrations determined in the field were consistent with the laboratory findings of others. In all instances, total residual chlorine concentrations less than 0.1 mg/l were found toxic to rainbow trout. Table 13 presents a summary of our findings as compared to toxicities shown by others. Charlotte and West Branch rainbow trout toxic concentrations are of the same order but lower than concentrations reported by Westfall, Coventry, Shelford and Miller (1935) Merkens (1958) and Washington Department of Fisheries (1960). Additional stress may have been placed on the fish by testing the two species together and having to maintain position in the river current. Other possible factors causing a difference in results were the characteristics of the diluent water, acclimation times, exposure times, and methods of total residual chlorine measurement in the various studies.

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Table 12. Summary of bioassay results obtained below four Michigan WWTP's.

Species-Plant	Effluent			Last Station Exhibiting Apparent Mortality[1]			Next Station Exhibiting No Apparent Mortality		
	Mean Total Chlorine Residual (mg/l)	Percent of River	Downstream Distance When Mixed (feet)	Average Survival (percent)	Distance from Outfall (feet)	Average Total Residual Chlorine Concentration (mg/l)	Average Survival (percent)	Distance from Outfall (feet)	Average Total Residual Chlorine Concentration (mg/l)
Rainbow Trout									
Mason	2.64	5.00	750	35	4,200	0.046	90	7,900	0.013
Charlotte	1.77	3.34	500	5	3,200	0.007 (0.02)[2]	90	10,600	0.000
West Branch	1.35	1.50	500	50	500	0.014	100	3,200	0.002
Roscommon	18.92	0.06	200	--	---	----	---	---	---
Fathead Minnow									
Charlotte	1.77	3.34	500	12.5	3,200	0.007 (0.02)[2]	65	10,600	0.000
Mason	2.64	5.00	750	30	750	0.072	75	4,200	0.046

(1) The last station exhibiting mortality that was obviously greater than natural or random mortality. This was mortality attributable to chlorinated compounds present in the municipal WWTP wastes.

(2) Average value with "zero" values not included.

Table 13. Toxic total residual chlorine concentrations below three Michigan WWTP's and toxicities reported by other authors.

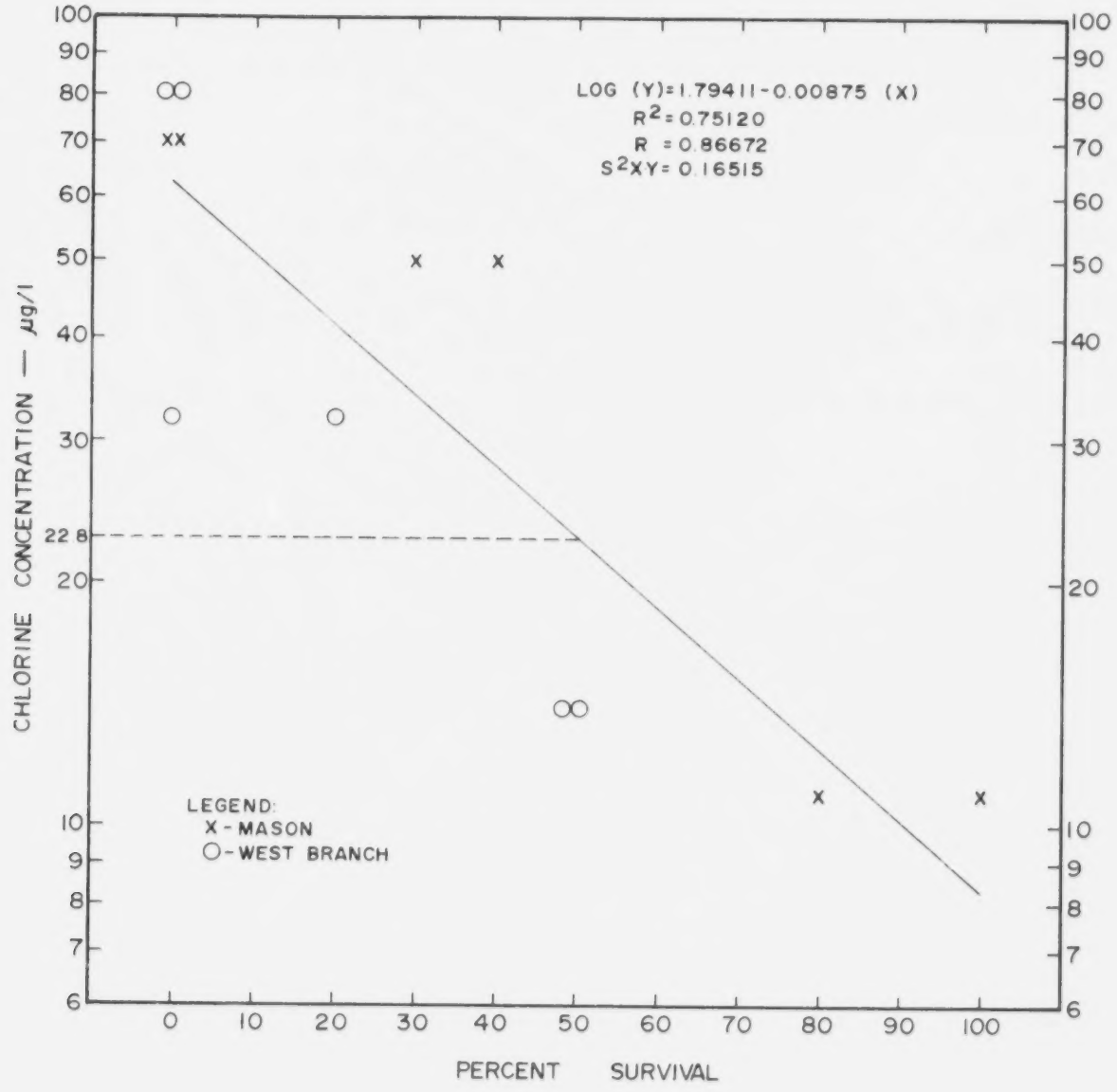
Study Site or Author	Lethal Level (mg/l)	Test Fish	Method of Chlorine Determination	Remarks
Charlotte.	0.007 (0.02)[1]	Rainbow Trout	A. T.	120 hour exposure.
West Branch.	0.014	Rainbow Trout	A. T.	96 hour exposure, 96 hour TL-50-0.014 mg/l.
Mason.	0.046	Rainbow Trout	A. T.	96 hour exposure, TL-50-0.029 mg/l.
Westfall	0.06	Trout Fry	Not Given	Cited in McKee & Wolf. (1963)
Coventry, Shelford & Miller (1935)	0.05	Trout Fry	Ortho-tolidine	Killed all trout after 48 hour exposure. 0.03 ppm instantly fatal to trout.
Merkens (1958) . . .	0.08	Rainbow Trout	A. T.	Killed half of fish after 7 day exposure.
Washington Dept. of Fisheries (1960)	0.1	Chinook, Pike & Silver Salmon in Fresh & Salt Water	Not Given	Critical level for 72 hour exposure, chloramines not formed in salt water under their conditions. Cited in McKee & Wolf. (1963)
Charlotte.	0.007 (0.02)[1]	Fathead Minnows	A.T.	120 hour exposure.
Mason.	0.072	Fathead Minnows	A.T.	96 hour exposure.
Zillich (1969 b) . .	0.05-0.016	Fathead Minnows	I.S.E.	96 hour TL-50 on-site bioassay trailer set up. Lethal threshold 0.04 mg/l.
Zillich (1969 c) . .	0.08-0.19	Fathead Minnows	I.S.E.	96 hour exposure on-site bioassay set-up. Lethal threshold 0.05 mg/l.
Arthur & Eaton . . . (1971)	0.085-0.154	Fathead Minnows	A.T.	96 hour TLM.

[1]Average value with "zero" values not included.
A.T. - Amperometric titration.
I.S.E. - Iodometric starch-iodide endpoint procedure given in Standard Methods.

In all instances the toxic concentrations for fathead minnows were less than 0.2 mg/l. Toxic concentration for fathead minnows at Charlotte were lower than those found by Zillich (1969 b and c) and Arthur and Eaton (1971). The Mason minnow results agree with these authors. The fathead minnow survival in these studies was highly variable. Rainbow trout appeared to survive better than fathead minnows did under both chlorinated and non-chlorinated exposures. A correlation coefficient was computed between the survival of rainbow trout and fathead minnows at the control stations for all four studies combined. An interaction or competition between species should result in a significant negative correlation between survival of these two species. The correlation coefficient computed was -0.02 indicating no significant correlation between the survival of these two species. A fungal infection, observed on some fathead minnows, seems the most probable cause of this difference in survival.

A generalized relationship between total residual chlorine concentration and rainbow trout survival at Mason and West Branch was computed by linear regression (Figure 9). This combined regression resulted in an r of 0.86672 and r^2 of 0.75120. The r value was significant beyond the 0.01 level. Approximately 75 percent of the data variance is due to the regression of these two variables. The computed 96-hour TL-50 concentrations was approximately 0.023 mg/l.

Figure 9. Chlorine concentration and regression for percent survival of rainbow trout below Mason and West Branch WWTP's outfall.



SECTION X

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SECTION XI

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SECTION XII

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Appendix II. Shellfish sampling data and descriptive statistics.

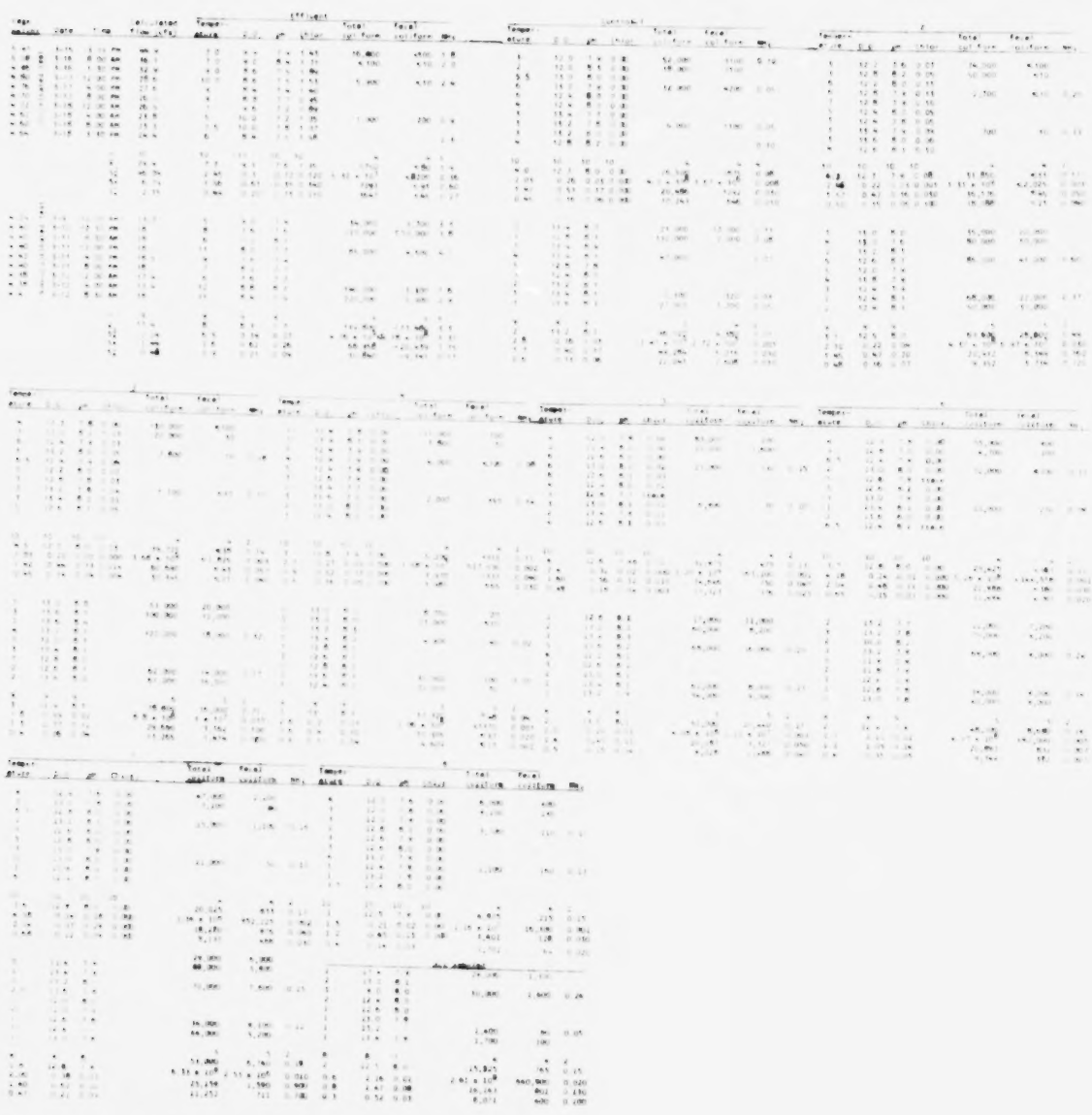
Date	Time	Calculated flow-rate	Time	Effluent Temperature	D.O.	pH	Chlor.	Total coliform	Fecal coliform	NH$_3$	Control Temperature	D.O.	pH	Chlor.	Total coliform	Fecal coliform	NH$_3$
2-11			11:00 AM	7.5	8.4	7.2	1.41				1.0	10.0	8.4	0.00			
2-11		40	4:00 AM	7.0	8.4	7.6	1.41				1.0	11.4	8.1	0.00			
2-11		40	2:30 PM	8.0	7.5	7.5	1.41	100	< 10	<2.0	1.0	12.4	8.1	0.00	14,000	140	0.27
2-11		40	4:00 PM	11.0	7.4	7.6	1.14			12.0	1.0	11.8	8.1	0.00			0.43
2-11		40	7:00 PM	11.0	7.4	7.6	1.14			12.0	1.0	11.8	8.1	0.00			0.43
2-12		41	12:00 AM	10.0	7.4	7.6	0.48	800	< 10	27.0	1.0	11.0	7.9	0.00			0.47
2-12		41	5:30 AM	11.0	7.0	7.6	1.41			29.0	1.0	11.4	7.8	0.00	23,000	110	0.25
2-12		34	12:00 PM	9.0	7.0	7.7	1.41	500	< 10	29.0	1.0	11.4	7.5	0.00			0.45
2-12		34	4:30 PM	9.0	7.0	7.7	1.53			26.0	1.0	11.4	7.8	0.00	11,000	140	0.31
2-13		44	11:30 AM	9.0	8.0	7.6	1.86	100	< 10	40.0	1.0	11.8	7.8	0.00			0.44
2-13		44	11:45 AM	8.0	7.0	7.6	1.65	100	< 10	34.0	1.0	12.0	7.7	0.00	13,000	80	0.44
2-13		44	8:30 PM	7.0	7.4	7.7	1.82	100	< 10	34.0	1.0	11.8	7.8	0.00	17,000	130	0.44
	11			11	11	11	11	7	6	10	11	11	11	11	7	6	10
	24			8.4	7.6	7.7	1.77	2,700	< 10	30.0	1.0	11.4	7.8	0.00	16,400	120	0.41
	14.4			0.88	0.54	0.14	0.431	2,700	0	30.0	0	0.55	0.23	0	4,300	17	0.09
	4.4			1.6m	0.49	0.14	0.431	< 91	0	3.0	0	0.55	0.23	0	12,900	51	0.09
	3.1			0.47	0.14	0.08	0.160	< 111	0	1.40	0.04	0.23	0.15	0	4,420	70	0.04
2-14		42	1:00 PM	8.4	7.2	7.7	1.77	1,300,000	140,000	24.0							
2-14		42	12:30 PM	9.0	7.2	7.6	40.0	900,000	900,000	25.0	1.0	11.4	7.7	0.0	140,000	400	0.34
2-14		42	4:00 PM	9.0	7.2	7.6	40.0				1.0	11.4	7.7	0.0	1,100	230	0.33
2-14		40	1:00 AM	8.0	7.2	7.6	40.0	1,400,000	180,000	23.0	2.0	11.4	7.7	0.0	2,100	130	0.32
2-14		39	4:00 AM	8.5	7.4	7.6					2.0	11.2	7.7	0.0			
2-14		43	8:00 AM	8.5	7.4	7.7					1.5	11.2	7.8				
2-14		40	12:00 PM	8.5	7.4	7.6	73.0	900,000	500,000	33.0	1.0	11.4	7.8				
2-14		41	12:00 PM	10.5	7.4	7.6	61.0	1,500,000	46,000	25.0	0.5	11.6	7.7	0.0	1,400	100	0.31
		41	12:00 PM	9.5	7.4	7.6	67.0	900,000	180,000	29.0	0.5	11.6	7.8	0.0	1,400	130	0.41
											0.5	11.6	7.8	0.0	1,000	400	0.37
	4	41		4	4	4	4	1,450,000	4	4	4	4	4	4	4	4	4
	4	41		8.9	7.3	7.6	64.0	2.47×10^{6}	495,000	26.8	1.0	11.4	7.8	0.0	1,200	80	0.36
	141.6			0.78	0.41	0.0	48.0	1.42×10^{6}	2.69×10^{6}	12.92	0.5	0.22	0.14	0.0	1.23×10^{6}	1.23×10^{6}	0.04
	11.6			0.27	0.11	0.0	0.0	57,643	165,000	1.60	0.5	0.37	0.07	0.0	1,468	1.2	0.04
	4.4			0.12	0.04	0.0	0.0		470,000	1.50	0.13	0.19	0.05	0.0	439,548	1.1	0.07

Temperature	D.O.	pH	Chlor.	Total coliform	Fecal coliform	NH$_3$
1.0	10.0	7.4	0.00			
1.0	9.8	7.4	0.00			
1.0	8.4	7.5	0.00			
1.0	8.4	7.6	1.03	< 100	< 10	
1.0	8.4	7.6	1.14			
1.0	10.0	7.4	1.04			
1.0	11.6	7.6	0.00	500	210	
1.0	10.4	7.4	0.00			
1.0	11.2	7.6	0.00	17,000	< 80	
1.0	11.2	7.6	0.00			
1.0	10.2	7.6	0.01			
1.0	8.4	7.6	1.11	< 100	10	
1.0	10.4	7.6	1.11	< 100	< 10	
1.0	10.0	7.4	0.24	< 100	< 10	
1	1	1	1	1	1	1
1.0	10.2	7.2	0.00	5,000	50	
1.0	10.2	7.6	0.00			
1.0	10.2	7.6	0.00	17,000	130	
1.0	10.2	7.6	0.00			
1.0	10.2	7.6	0.00			
1.0	10.2	7.6	0.00	

APPENDIX 2

Charlotte Wastewater Treatment Plant
Final Effluent Discharge Data

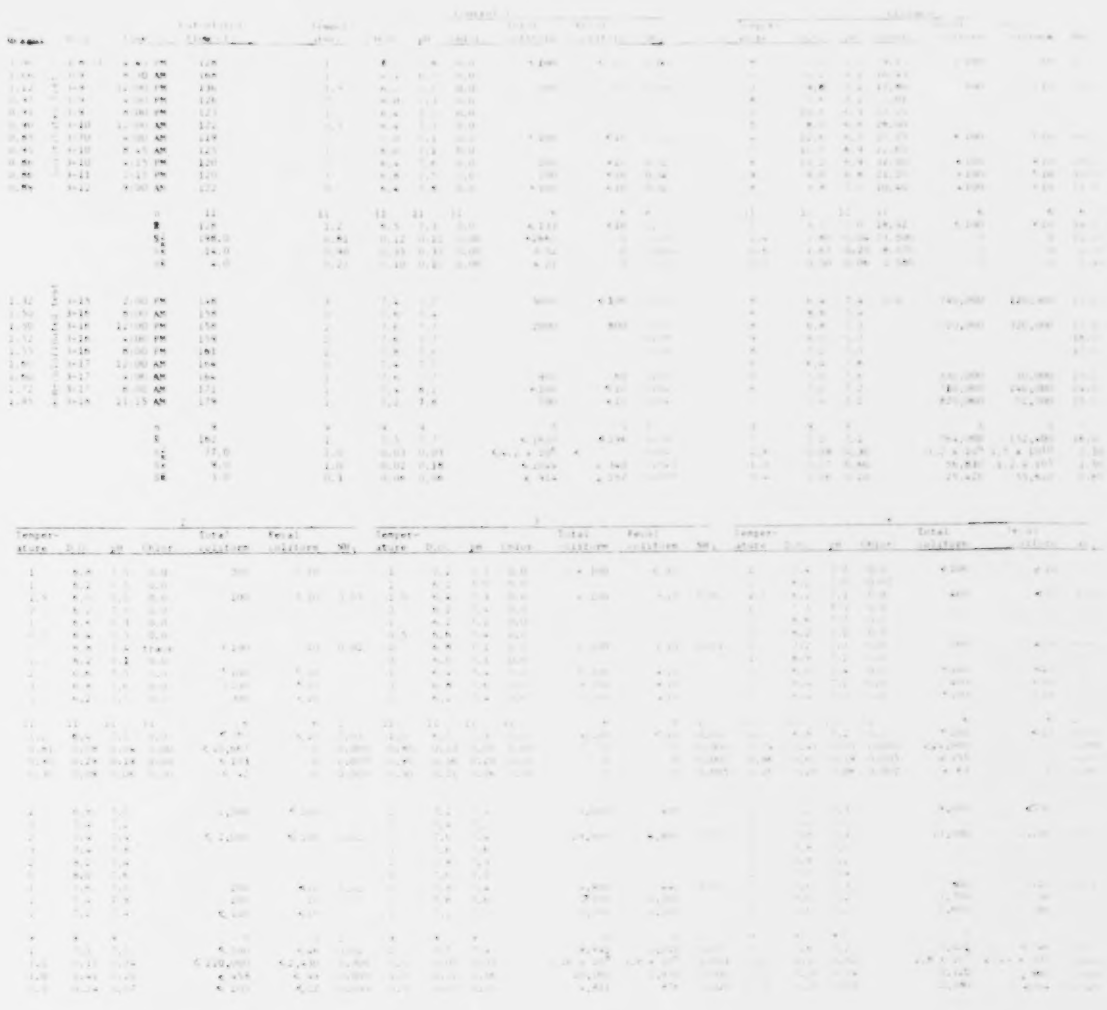
Date	Flow-cfs	BOD-mg/l	Suspended Solids-mg/l	Suspended Volatile Solids-mg/l	Weather High Temp°F	Weather Low Temp°F	Precipitation-inches	
2- 1-71	0.6909	125	78	54	8	-16	0.02	
2- 2-71	0.6641				13	-17	0.00	
2- 3-71	0.5731	90	64	54	29	8	0.11	
2- 4-71	1.0937				33	21	0.20	
2- 5-71	0.8449	89	82	74	47	23	0.90	
2- 6-71	0.7241				27	18	0.00	
2- 7-71	0.5967				26	5	0.00	
2- 8-71	0.6527	89	70	62	21	5	0.00	
2- 9-71	0.6106				17	1	0.01	
2-10-71	0.5790	97	74	70	23	7	0.00	Chlorinated Phase
2-11-71	0.6427				42	6	0.00	
2-12-71	0.5903	86	54	38	39	20	0.17	
2-13-71	0.6702				21	5	0.00	
2-14-71	0.5350				23	-5	0.00	
2-15-71	0.7024	88	62	52	31	19	0.02	
2-16-71	0.6407				35	22	0.00	
2-17-71	0.7555	98	76	60	41	28	0.00	
2-18-71	1.2083				47	32	0.15	
2-19-71	1.7639	67	58	50	46	35	0.64	
2-20-71	2.3297				47	30	0.09	
2-21-71	2.0272				32	27	0.00	
2-22-71	1.4876	101	76	66	33	25	0.11	
2-23-71	0.7516				34	28	0.15	
2-24-71	0.6831	78	44	38	38	16	0.00	
2-25-71	0.8019				49	23	0.00	
2-26-71	0.8699	82	78	68	51	32	0.00	
2-27-71	0.8689				51	31	0.07	
2-28-71	0.7140				37	28	0.00	
n	28	12	12	12				
$\bar{x}$	0.8955	91	68	57				
s_x^2	0.21140	200.9	135	137.0				
$s_{\bar{x}}$	0.45980	14.2	11.6	11.7				
$s_{\bar{x}}^2$	0.08690	4.1	3.4	3.4				
3- 1-71	0.7394	88	68	62	39	18	0.00	
3- 2-71	0.7140				34	18	0.00	
3- 3-71	0.6245	82	52	38	29	16	0.00	
3- 4-71	0.7221				37	9	0.00	
3- 5-71	0.8291	83	56	46	46	21	0.00	Non Chlorinated Phase
3- 6-71	0.7817				39	33	0.46	
3- 7-71	0.6209				33	23	0.31	
3- 8-71	0.7291	86	68	60	25	17	0.00	
3- 9-71	0.6858				33	-2	0.00	
3-10-71	0.6367	90	66	58	34	26	0.21	
3-11-71	0.6539				38	21	0.02	
3-12-71	0.7112	90	92	82	36	16	0.02	
3-13-71	0.7527				39	30	0.00	
3-14-71	1.0030				65	33	0.00	
3-15-71	1.0783	90	76	58	59	36	0.17	
3-16-71	0.9976				39	25	0.03	
3-17-71	0.7088	93	74	58	38	16	0.00	
3-18-71	0.7116				39	18	0.02	
3-19-71	0.7997	81	48	42	37	30	0.31	
3-20-71	0.7840				34	27	0.02	
3-21-71	0.6872				43	16	0.00	
3-22-71	0.8344	97	48	42	39	20	Trace	
3-23-71	0.7371				31	20	0.02	
3-24-71	0.6884	93	70	66	30	11	0.06	
3-25-71	0.7018				35	7	0.00	
3-26-71	0.6983	86	44	36	41	20	0.00	
3-27-71	0.7137				47	26	0.14	
3-28-71	0.6888				44	32	0.00	
3-29-71	0.7737	90	66	52	40	29	0.00	
3-30-71	0.7436				47	17	0.00	
3-31-71	0.7134	86	92	68	65	25	0.00	
n	31	14	14	14				
$\bar{x}$	0.7512	87	64	55				
s_x^2	0.01210	65.3	267.2	169.1				
$s_{\bar{x}}$	0.11020	8.1	16.3	13.0				
$s_{\bar{x}}^2$	0.01980	2.2	4.4	3.5				



APPENDIX 4

West Branch Wastewater Treatment Plant
Final Effluent Discharge Data

	Date	Flow-cfs	BOD-mg/l	Suspended Solids-mg/l	Suspended Volatile Solids-mg/l	Weather High Temp.°F	Weather Low Temp.°F
	3- 1-71	0.256				36	-1
	3- 2-71	0.253	24	32	36	38	0
	3- 3-71	0.244				26	3
	3- 4-71	0.239	16	37	35	34	8
	3- 5-71	0.232				43	18
	3- 6-71	0.223	24	43	36	36	14
	3- 7-71	0.227				30	10
	3- 8-71	0.239				27	3
Non-chlorinated phase	3- 9-71	0.229	26	19	22	42	10
	3-10-71	0.224				36	5
	3-11-71	0.223	3	13	20	53	6
	3-12-71	0.228				52	16
	3-13-71	0.224	23	19	28	48	24
	3-14-71	0.284				53	31
	3-15-71	0.321				43	20
Chlorinated phase	3-16-71	0.287		31	35	33	10
	3-17-71	0.281				39	10
	3-18-71	0.275	50	57	37	41	17
	3-19-71	0.270				--	--
	3-20-71	0.231	26	36	41	37	12
	3-21-71	0.247				53	12
	3-22-71	0.252				53	13
	n	22	8	9	9		
	$\bar{x}$	0.250	24	32	32		
	$s_{\bar{x}}^2$	0.0006	170.0	185.9	51.9		
	s_x	0.0244	13.0	13.6	7.2		
	$s_{\bar{x}}$	0.0052	4.6	4.5	2.4		



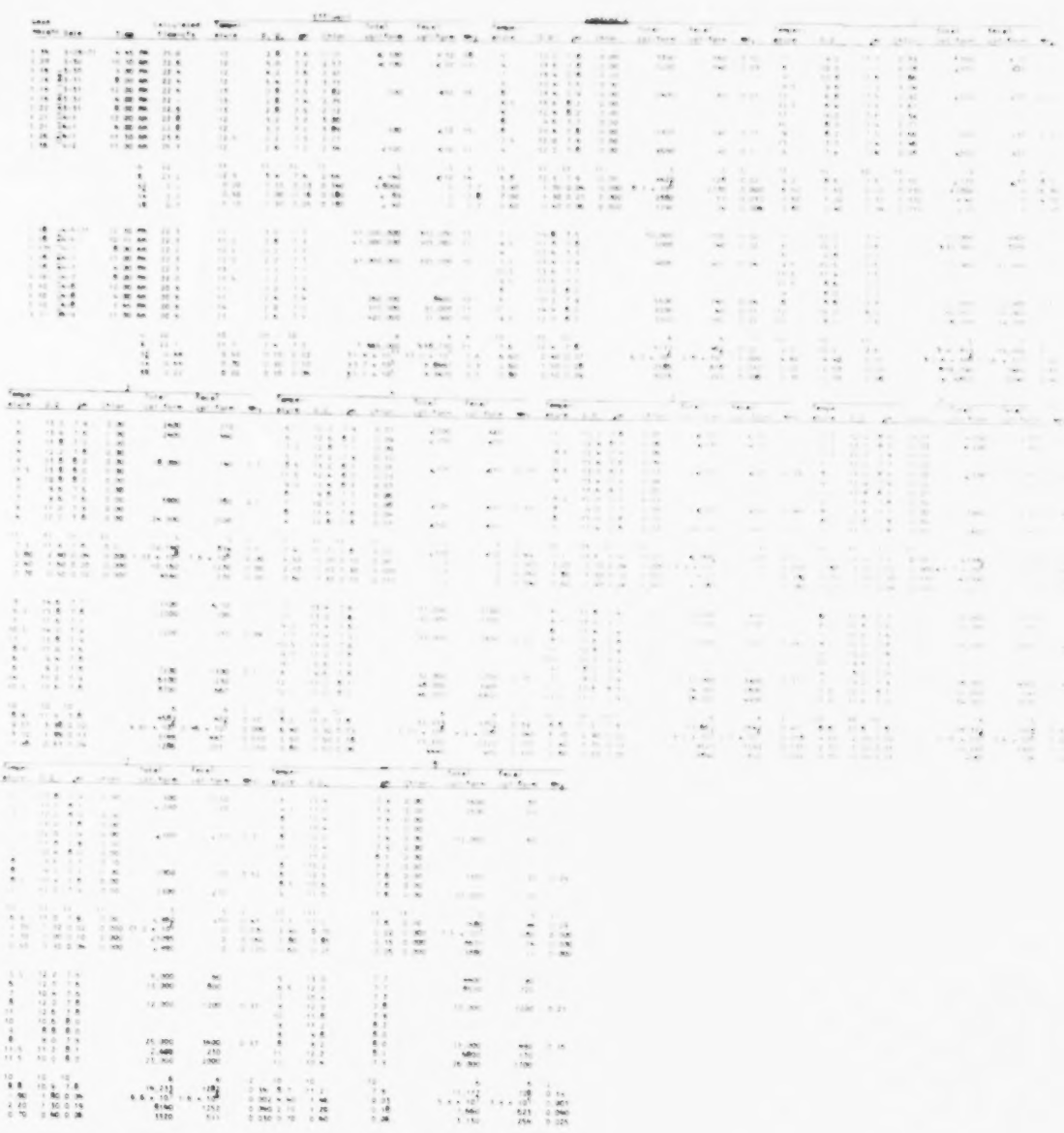
Note: D.O. (dissolved oxygen), Chlor. (total chlorine residual) and NH_3 (ammonia as nitrogen) expressed as mg/l.
T (temperature) - degrees centigrade
Total coliform and fecal coliform - organisms/100 ml

APPENDIX 6

Roscommon Wastewater Treatment Plant
Final Effluent Discharge Data

Date		Flow	Weather: High Temp. °F	Weather: Low Temp. °F	Precipitation
3-1-71	Chlorinated phase	55286	40	0	
3-2-71		61530	32	-10	
3-3-71		48490	28	8	
3-4-71		64880	20	-3	
3-5-71		45680	34	0	
3-6-71		51420	42	30	0.2
3-7-71		56080	38	22	0.6
3-8-71		44980	20	8	
3-9-71		49060	19	10	
3-10-71		-----	30	10	
3-11-71		57240	32	0	
3-12-71		51140	39	3	
3-13-71		20830	40	10	
3-14-71		28070	42	32	
3-15-71	Non-chlorinated phase	44060	50	42	
3-16-71		92040	40	20	
3-17-71		39040	38	11	
3-18-71		65460	40	19	
3-19-71		40080	42	16	
3-20-71		50600	34	27	
3-21-71		22400	32	15	
3-22-71		27050	40	3	
3-23-71		49310	40	20	
3-24-71		43870	35	10	
3-25-71		40750	20	-10	

n	24
$\bar{x}$	47889
s_x^2	2.36×10^8
s_x	15350
$s_{\bar{x}}$	3133



Note: D.O. (dissolved oxygen), Chlor. (Total chlorine residual) and NH_3 (ammonia as nitrogen, expressed as mg/l).
T (temperature), Degrees Centigrade.
Total coliform and fecal coliform - organisms/100 ml.

APPENDIX 8

Mason Wastewater Treatment Plant
Final Effluent Discharge Data

					Weather		
	Flow	BOD	Suspended	Suspended Volatile		High	Low
Date	MGD	mg/l	Solids-mg/l	Solids-mg/l	Precipitation-in	Temp.°F	Temp.°F
3- 1-71	0.730	6	13	9		38	19
3- 2-71	0.716	17	13	9		32	18
3- 3-71	0.718	8	10	8		23	12
3- 4-71	0.694	10	16	8		36	15
3- 5-71	0.748	21	30	24	0.12	44	30
3- 6-71	0.845				0.54	36	25
3- 7-71	0.571				0.15	30	19
3- 8-71	0.720	9	17	14		25	6
3- 9-71	0.699	6	18	12	0.09	33	13
3-10-71	0.679	6	11	7	0.10	30	25
3-11-71	0.745					34	19
3-12-71	0.779	13	22	10	0.02	33	29
3-13-71	0.682					44	32
3-14-71	1.644					63	44
3-15-71	1.368	15	21	19	0.25	42	28
3-16-71	1.032	9	14	11		30	21
3-17-71	0.971	9	19	14		35	21
3-18-71	1.001	11	15	10	0.20	38	27
3-19-71	1.140	14	16	14	0.15	36	30
3-20-71	0.923					32	20
3-21-71	0.690					40	20
3-22-71	1.004	13	15	11	0.01	39	23
3-23-71	0.925	10	14	10		30	16
3-24-71	0.916	8	12	9	0.04	29	9
3-25-71	0.893					36	18
3-26-71	0.944					41	28
3-27-71	0.793					44	34
3-28-71	0.639					43	35
3-29-71	0.885	20	33	21		36	22
3-30-71	0.848					45	28
3-31-71	0.833	23	20	15		62	37
n	31	19	19	19			
$\bar{x}$	0.864	12	17	12			
s_x^2	0.0482	26.8	36.0	21.7			
s_x	0.2195	5.2	6.0	4.7			
$s_{\bar{x}}$	0.0394	1.2	1.4	1.1			
4- 1-71	0.862	15	16	9	0.06	54	31
4- 2-71	0.882	11	10	10	0.05	38	23
4- 3-71	0.659					26	19
4- 4-71	0.718					36	20
4- 5-71	0.812	8	9	9		44	22
4- 6-71	0.801	12	6	6		51	23
4- 7-71	0.757	9	5	2		58	31
4- 8-71	0.793	10	10	7		67	51
4- 9-71	0.808				Trace	64	26
4-10-71	0.607					60	42
4-11-71	0.595					70	63
4-12-71	0.891	7	9	7	0.49	77	46
4-13-71	0.876	7	7	6	0.08	48	27
4-14-71	0.836	6	16	9		48	30
4-15-71	0.855					60	34
4-16-71	0.832	9	14	7	0.03	70	48
4-17-71	0.758					62	48
4-18-71	0.680					62	33
4-19-71	0.727					68	37
4-20-71	0.806					74	43
4-21-71	0.782					52	40
4-22-71	0.776	29	26	21		59	31
4-23-71	0.720	27	23	18		61	28
4-24-71	0.714					48	30
4-25-71	0.777					52	36
4-26-71	0.758	11	18	6		50	29
4-27-71	0.738				0.05	45	41
4-28-71	0.681				0.06	49	39
4-29-71	0.677	16	8	8		48	28
4-30-71	0.726	18	12	10	0.03	56	38
n	30	15	15	15			
$\bar{x}$	0.764	13	13	9			
s_x^2	0.0059	47.0	38.3	22.6			
s_x	0.0768	7.0	6.2	4.8			
$s_{\bar{x}}$	0.0140	1.8	1.6	1.2			

Chlorinated Phase

Non-Chlorinated Phase

1 Accession Number: W

2 Subject Field & Group:

SELECTED WATER RESOURCES ABSTRACTS
INPUT TRANSACTION FORM

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23 Descriptors (Starred First):
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25 Identifiers (Starred First):
*Toxicity, *Chlorinated Municipal Waste, Fish

27 Abstract: This project consisted of separate studies at four different Michigan municipal wastewater treatment plants. Ten rainbow trout (Salmo gairdneri) and ten fathead minnows (Pimephales promelas), each previously acclimated to the river, were held for 96 hours in live boxes in the receiving stream above and below these plant outfalls. Fish held below these outfalls were subjected to both chlorinated and non-chlorinated exposures during effluent discharge. During fish exposure, the test waters were monitored chemically and bacteriologically.

Total residual chlorine concentrations below three of the four plants were toxic to rainbow trout at distances up to 0.8 mile. Fathead minnows appeared adversely affected up to 0.6 mile downstream in two of four plants. Total residual chlorine concentrations less than 0.1 mg/l were toxic to fathead minnows in the plants.

The rainbow trout 96-hour total residual chlorine TL-50 concentration below two plants was 0.023 mg/l.

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